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INTERNATIONAL APPLICATION NO.
PCT/KR99/00687

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TITLE OF INVENTION **BIOCIDE COMPOSITION AND STERLIZATION METHOD USING THE SAME**

APPLICANT(S) FOR DO/EO/US **Ki-Seung Choi, Jin-Man Kim, Jeong-Ho Park, Myung-Ho Cho and Soon-Jong Hahn**

Applicant herewith submits to the United States Designated /Elected Office (DO/EO/US) the following items and other information:

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☒ This express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and PCT Articles 22 and 39(I).
4. ☐ A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. ☐ A copy of the International Application as filed (35 U.S.C. 371(c)(2))
 - a. ☐ is transmitted herewith (required only if not transmitted by the International Bureau).
 - b. ☐ has been transmitted by the International Bureau.
 - c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☐ A translation of the International Application into English (35 U.S.C. 371(c)(2)).
7. ☐ A copy of the International Search Report (PCT/ISA/210)
 - a. ☐ are transmitted herewith (required only if not transmitted by the International Bureau).
 - b. ☐ have been transmitted by the International Bureau
 - c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
 - d. ☐ have not been made and will not be made.
8. ☐ A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ☐ An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. ☐ A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

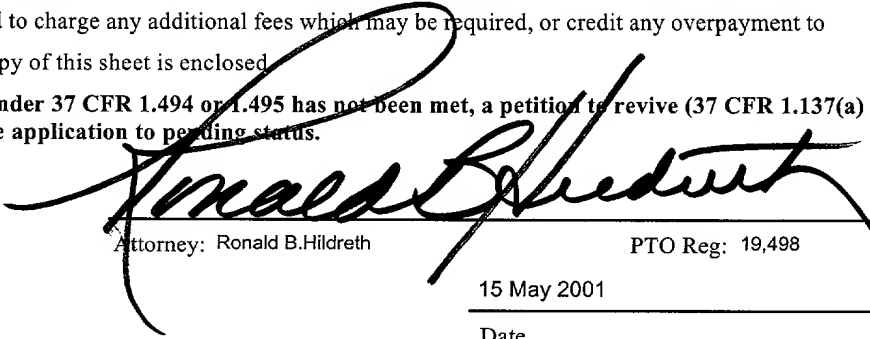
Items 11. to 16. below concern other document(s) or information included:

11. ☐ A copy of the International Preliminary Examination Report (PCT/IPEA/409)
12. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. ☐ A **FIRST** preliminary amendment.
☐ A **SECOND** or **SUBSEQUENT** preliminary amendment.
14. ☐ A substitute specification.
15. ☐ A change of power of attorney and/or address letter.
16. ☒ Other items or information:

- a. ☒ a copy of the International Search Report (PCT/ISA/210)
- b. ☐ a copy of the International Preliminary Examination Report (PCT/IPEA/409)

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09/831929 INTERNATIONAL APPLICATION NO. PCT/KR99/00687		INTERNATIONAL FILING DATE 16 NOVEMBER 1999		PRIORITY DATE CLAIMED 16 NOVEMBER 1998	
17. <input checked="" type="checkbox"/> The following fees are submitted: Basic National Fee (37 CFR 1.492(a)(1)-(5): Neither international preliminary examination fee (37 CFR 1.482) Nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO (1.492(a)(3)) \$1,000.00 International preliminary examination fee (37 CFR 1.482) not paid to USPTO but International Search Report prepared by the EPO or JPO (1.492(a)(5)) \$860.00 International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1.445(a)(2)) paid to USPTO (1.492(a)(2)) \$710.00 International preliminary examination fee paid to USPTO (37 CFR 1.482) but all claims did not satisfy provisions of PCT Article 33(1)-(4) (1.492(a)(1)) \$690.00 International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(1)-(4) \$100.00 <div style="text-align: right;">ENTER APPROPRIATE BASIC FEE AMOUNT = \$ 1,000</div>				CALCULATIONS <small>PTO USE ONLY</small>	
Surcharge of \$130.00 for furnishing the oath or declaration later than <input type="checkbox"/> 20 <input type="checkbox"/> 30 months from the earliest claimed priority date (37 C.F.R. 1.492)(e)).				\$	
Claims	Number Filed	Number Extra	Rate	\$	
Total Claims	5 -20=	0	X \$ 18.00	\$	0
Independent Claims	2 -3=	0	X \$ 80.00	\$	0
Multiple dependent claim(s) (if applicable)			+ \$270.00	\$	
TOTAL OF ABOVE CALCULATIONS =				\$	1,000
Reduction by 1/2 for filing by small entity, if applicable.				\$	
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TOTAL NATIONAL FEE =				\$	1,000
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). \$40.00 per property				\$	
TOTAL FEES ENCLOSED =				\$	1,000
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a. <input checked="" type="checkbox"/> A check in the amount of \$ <u>1,000</u> to cover the above fees is enclosed. b. <input type="checkbox"/> Please charge our Deposit Account No. <u>02-4377</u> in amount of \$ _____ to cover the above fees. A copy of this sheet is enclosed. c. <input checked="" type="checkbox"/> The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. <u>02-4377</u> . A copy of this sheet is enclosed.					
NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.					
SEND ALL CORRESPONDENCE TO: Ronald B. Hildreth BAKER BOTTS L.L.P. 30 Rockefeller Plaza New York, New York 10112-4498					
 Attorney: Ronald B. Hildreth				PTO Reg: 19,498	
				15 May 2001	
				Date	

BIOCIDE COMPOSITION AND STERILIZATION METHOD USING THE SAME

This application is based on application No. 98-49095 filed in the Korean Industrial Property Office on November 16, 1998, the contents of which are
5 incorporated hereinto by reference.

BACKGROUND OF THE INVENTION**(a) Field of the Invention**

The present invention relates to a biocide composition, more particularly to a biocide composition having synergistic effects by comprising 3-isothiazolone
10 and polyhexamethyleneguanidine phosphate and a method for sterilizing microorganisms, fungi, etc. using the same.

(b) Description of the Related Art

Microorganisms, bacteria, mold, algae, etc. grow in industrial water such
15 as cooling water, wastewater, emulsifiers used in the textile industry, etc. and have a deteriorating effective on the operations of industrial processes. Such microorganisms propagate using organisms contained in the industrial water as a nutrient source and secrete polysaccharides. Varieties of organic and inorganic substances combined with these secreted polysaccharides and form
20 viscous lumps or masses also called slime. Organic materials such as cellulose, hemicellulose, and the fibrin of white water in paper industry in particular provide to have sufficient nutrient sources for such microorganisms. The slime formed at areas of low fluid flow in a paper processing process results in both direct and indirect losses, such as manufacturing time loss, equipment
25 efficiency deterioration, etc. due to the deterioration of pulp quality, etc. Furthermore, the growth of microorganisms at such places like a cooling water facility, where much water is contained or recirculated causes a fouling phenomena. This deteriorates heat transfer efficiency in an industrial cooling tower, as well as corrodes metal or erodes wooden parts..

30 Bacteria such as taloblastic prokaryotes can propagate by decomposing

various types of organisms, some of which are the sources of the microbilogically induced corrosion, by secreting and discharging polysaccharides forming a biofilm. Fungi such aseukaryote can also propagate by decomposing various types of organisms like bacteria, and some types of
5 fungi secreting cellulase are the sources for discoloration and decomposition of wooden parts by way of degrading the fibroid materials of wooden parts of a cooling tower, etc.

Algae, such as eukaryote, can propagate by photosynthesizing under an environment with light, air, and a small amount of organisms, and carbohydrate
10 formed from algae is used as a nutrient source for other microorganisms such as bacteria and fungi, thereby accelerating the fouling phenomena. Algal fouling due to algae propagation is intensified at places that are exposed to sunlight, particularly in cooling water facilities, swimming pools, etc. This phenomena results in clogging which blocks water pipes, as well as the
15 deterioration of heat transfer efficiency, oxidization of metal surfaces by the generation of oxygen, and the promotion of corrosion at a holes on metallic surfaces through a partial galvanic reaction when the organism dies.

Various biocides are being developed in order to kill such microorganisms, fungi, algae, etc. or to prevent their surface adhesion to metal,
20 etc. These biocides are generally divided into oxidant biocides and non-oxidant biocides. The oxidant biocides that are mainly used are halogen compounds such as chlorine, bromine, etc. that are popular due to their economic advantage of strong oxidation capability and low price. However, they can cause erosion of the wooden parts of cooling towers and metallic decomposition, and their
25 sterilizing efficiency tends to deteriorate as they can be easily being discharged into atmosphere. Furthermore, their practical effectiveness is poor due their peculiar way of first reacting with secreted polysaccharides before they react with microorganisms that are the actual sources of biofilm formation.

Non-oxidant biocides which overcome these disadvantages include 3-
30 isothiazolone, quaternary ammonium salt, formaldehyde emission compound, glutaraldehyde, etc. and are mainly used separately. Although 3-isothiazolone, which is disclosed in United States Patent Nos. 3,761,488, 4,105,431, 4,279,762, etc., has a high sterilizing effect and wide antibiotic spectrum, it has a

disadvantage in that its immediate instantaneous sterilizing effects are low. Furthermore, Korean Patent Application No. 89-20381 discloses an antiseptic composition useful in preventing circulation water putrefaction wherein a biocides 5-chloro-2-methyl-4-isothiazolone-3-on and 2-methyl-4-isothiazolone-3-on are mixed in a ratio of about 3:1 and wherein this antiseptic composition further comprises didecyldimethylammoniumchloride. United States Patent No. 4,379,137 discloses a method for improving sterilizing capability by mixing polymer quaternary ammonium salt and 3-isothiazolone. However, since these mixtures emit corrosive materials, i.e., halogen compounds such as fluorine, chlorine, etc., it is difficult for them to be applied where metals susceptible to corrosion are used, such as carbon steel, cast iron, stainless steel, copper, etc.

Additionally, Korean Patent Application No. 97-80170 discloses a process that can be applied even when metals susceptible to corrosion are used since the disclosed process does not emit a halogen compound. Another water soluble biocide composition having synergistic effects comprising 3-isothiazolone, which has high sterilizing effect on quaternary ammonium phosphate and microorganisms, is disclosed in Korean Patent Application No. 97-46517 as a biocide having superior properties of immediate sterilizing effects on microorganisms, durability, anticorrosiveness, etc. However, there is a problem in applying this invention to various industrial fields due to the issues such as bubbling or foaming, etc. when quaternary ammonium is added to provide immediate effectiveness to the 3-isothiazolone.

Furthermore, while polyhexamethyleneguanidine phosphate has immediate effectiveness, and is used in the effective and wide control of microorganisms in various industrial fields, including water treatment, as well as exhibiting low foaming properties, it has a disadvantage of not having a wide antibiotic spectrum by itself.

SUMMARY OF THE INVENTION

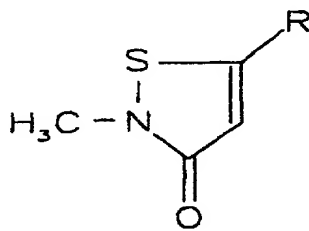
It is an object of the present invention to provide a biocide composition which can be used in the process where metals susceptible to corrosion are used, such as carbon steel, cast iron, stainless steel, copper, etc., as well as which has high sterilizing capability, a wide antibiotic spectrum, and superior

antiseptic effects.

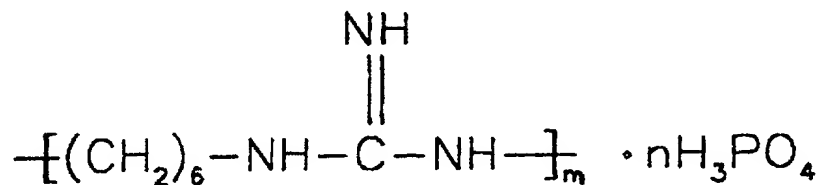
It is an other object of the present invention to provide a biocide composition which can not only be applied to various industrial processes on account of its low foaming features, but also which has a high sterilizing capability even when a small amount is used.

The present invention provides a biocide composition comprising 3-isothiazolone of the following General Formula 1 and polyhexamethyleneguanidine phosphate of the following General Formula 2 in order to accomplish the above objects:

10 [General Formula 1]



[General Formula 2]



15

where R is hydrogen or chlorine in the above General Formula 1 in the above General Formula 1, m is integer from 4 to 7, and n is an integer from 1 to 14 in the above General Formula 2. The present invention further provides a sterilizing method which kills or restrains the growth of bacteria, fungi or algae by inputting the above biocide composition into a media that is contaminated by

bacteria, fungi, and algae.

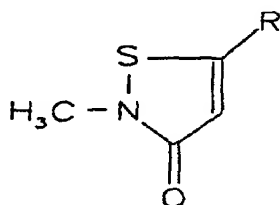
DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

In the following detailed description, only the preferred embodiments of the invention have been shown and described, simply by way of illustration of the best mode contemplated by the inventor(s) of carrying out the invention. As will be realized, the invention is capable of modification in various obvious respects, all without departing from the invention. Accordingly, the description is to be regarded as illustrative in nature, and not restrictive.

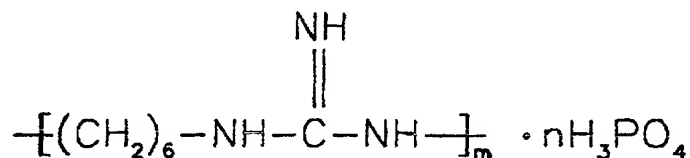
The present invention is described in detail below.

The present invention provides an antiseptic and/or biocide composition comprising 3-isothiazolone of the below General Formula 1 and polyhexamethyleneguanidine phosphate of the below General Formula 2:

[General Formula 1]



[General Formula 2]



where R is hydrogen or chlorine in the above General Formula 1 in the above General Formula 1, m is integer from 4 to 7, and n is an integer from 1 to 14 in the above General Formula 2.

In the above General Formulae, 3-isothiazolone is preferably 2-methyl-4-isothiazolone-3-one having R group of hydrogen or 5-chloro-2-methyl-4-

isothiazolone-3-on having R group of chlorine, and more preferably a mixture in which 2-methyl-4-isothiazolone-3-on and 5-chloro-2-methyl-4-isothiazolone-3-on are mixed in a weight ratio of 1:20 to 20:1. If the above ratio of 3-isothiazolone compound is out of the range of 1:20 to 20:1, the sterilizing effects are
5 deteriorated.

The mixing ratio of the above 3-isothiazolone and polyhexamethyleneguanidine phosphate is preferably a weight ratio of 1:1 to 1:65, more preferably 1:1 to 1:4. If the above mixing ratio of 3-isothiazolone and polyhexamethyleneguanidine phosphate is out of the above range, the
10 synergistic effects by the mixture of two biocides are deteriorated or not seen.

If the present invention of a mixture of 3-isothiazolone and polyhexamethyleneguanidine phosphate are used as a biocide composition, 3-isothiazolone compensates for the disadvantage in the low sterilizing effects of polyhexamethyleneguanidine phosphate on fungi and some bacteria, while
15 polyhexamethyleneguanidine phosphate compensates for the disadvantage of 3-isothiazolone has in its low initial and immediate sterilizing capability and effectiveness. Synergistic effects are seen in that the sterilizing action of a mixed biocide becomes greater than the sum of the sterilizing action that each component compound has. That is, the present invention of a composition can
20 restrain microorganisms more widely and effectively by mixing the two types of compounds having different sterilizing mechanisms, and obtains further effects such as the lowering of an emergence frequency of resistant strains more than in the case of using a single component compound.

A biocide composition of the present invention is preferably used in an
25 aqueous solution and more preferably used by putting 5 to 1,000 parts per million (ppm) of it into cooling water, etc. in which the microorganisms exist, although the amount used has no limit as long as it achieves the targetted sterilizing effects. A biocide composition of the present invention can be used in controlling microorganisms widely and effectively in the various industrial
30 fields such as in pulp and paper plants, cooling towers, as a disinfectant, etc.. In particular, it can be used by adding it to the cooling water of an industrial process, to disinfectant, paint, latex antiseptic, cosmetic additives, additives for emulsion products such as shampoo, etc., slime control chemicals for textile

weaving, paper slime control agent, and antiseptics for leather goods, metal processing oil, etc.

COMPARATIVE EXAMPLES for helping in gaining in an understanding
5 of the present invention and preferable EXAMPLES are described below.

[EXAMPLES 1-6]

The sterilizing effects of 3-isothiazolone and polyhexamethyleneguanidine phosphate on a mixed solution of 8 types of
10 bacteria (*Enterobacter aerogens* ATCC 13048, *Escherichia coli* ATCC 11229, *Micrococcus luteus* ATCC 9341, *Pseudomonas aeruginosa* ATCC 15442, *Klebsiella pneumoniae* ATCC 1560, *Staphylococcus epidermis* ATCC 155, *Staphylococcus aureus* ATCC 6538, and *Bacillus subtilis* ATCC 6984) were tested using Tryptic Soy Broth manufactured by Difco Corporation according to
15 the two fold dilution method. The minimum inhibiting concentration of each biocide and mixtures thereof were determined by observation with the naked eye of the degree of muddiness of the lowest concentration in which growth was deteriorated after culturing a culture fluid in which biocides and microorganisms are added at 30 degrees centigrade for 3 days.

20 It was determined that a synergistic effect of a biocide was seen when the sum of QA/Qa and QB/Qb was less than 1, as in the following EQUATION according to the method published in the paper of Kull, F. C. et al (*Appl. Microbiol.* 9:538~544 (1961)), and these results are represented in Table 1:

$$\text{Synergistic Index (SI)} = (QA/Qa) + (QB/Qb)$$

25 where Qa and Qb are MIC values (ppm) of a single compound A and single compound B, respectively, and QA and QB are MIC values (ppm) of the compounds A and B, respectively, out of each mixture.

[Table 1]

	Qa	Qb	QA	QB	QA/Qa	QB/Qb	SI
EXAMPLE 1	18.8	312.5	9.4	9.8	0.5	0.03	0.53
EXAMPLE 2	18.8	312.5	9.4	19.5	0.5	0.06	0.56
EXAMPLE 3	18.8	312.5	9.4	39.1	0.5	0.13	0.63

EXAMPLE 4	18.8	312.5	9.4	78.1	0.5	0.25	0.75
EXAMPLE 5	18.8	312.5	9.4	156.3	0.5	0.50	1.0
EXAMPLE 6	18.8	312.5	2.4	156.3	0.13	0.50	0.63

Qa : MIC value (ppm) on the mixed strain of a single 3-isothiazolone;

Qb : MIC value (ppm) on the mixed strain of a single poly-hexamethyleneguanidine phosphate;

5 QA : MIC value (ppm) of 3-isothiazolone out of a mixture;

QB : MIC value (ppm) of polyhexamethyleneguanidine out of a mixture.

As represented in the above Table 1, it can be seen that the same microorganism killing effect (SI was 0.53 in EXAMPLE 1) can be obtained even though only a half amount of 3-isothiazolone is used and the amount of
10 polyhexamethyleneguanidine phosphate used is reduced to 9.8 ppm. The same microorganism killing effect (SI was 0.63 in EXAMPLE 6) can also be obtained even though only a half amount of polyhexamethyleneguanidine phosphate is used and only 2.4 ppm of 3-isothiazolone is used. Therefore, it can be shown that a mixture of the above two compounds can inhibit the growth of bacteria
15 more effectively than the single use of each of the two compounds, and the synergistic action ratio of 3-isothiazolone and polyhexamethyleneguanidine phosphate is most preferable in the range of 1:1 to 1:4, where SI is from 0.53 to 0.63.

[TEST EXAMPLE 1 AND COMPARATIVE EXAMPLE 1]

20 The minimum inhibition concentration (MIC) values of a biocide composition in which 3-isothiazolone and polyhexamethyleneguanidine phosphate are mixed in a ratio of 1:4 (1 wt% of 3-isothiazolone and 15 wt% of 25 wt% poly-hexamethyleneguanidine phosphate) and of a single biocide of 1.5 wt% 3-isothiazolone were measured on 7 types of individual strains. After
25 diluting a biocide using 96 multi wall plates according to the two fold continuous dilution method, microorganisms were inoculated at a concentration of 104 CFU/ml. Then, after culturing at 30 degrees centigrade for 48 hours, the MIC values were measured by observation with the naked eye of the growth of microorganisms on the basis of muddiness. The results are represented in

Table 2.

The muddiness was observed using Tryptic Soy Broth manufactured by Difco Corporation as a medium in order to measure MIC values and strains of *Enterobacter aerogens* ATCC 13048, *Staphylococcus aureus* ATCC 6538, *Staphylococcus epidermis* ATCC 155, *Bacillus subtilis* ATCC 6984, *Saccharomyces cerevisiae* ATCC 9763, *Rhizopus oryzae* ATCC 10404, *Aspergillus niger* ATCC 9642 were used from among the strains which were used in EXAMPLE 1.

10

[Table 2]

MIC test results on microorganisms of a biocide mixture and a single biocide of 1.5 wt% 3-isothiazolone (units: ppm):

Strains Used	COMPARATIVE EXAMPLE (1.5% isothiazolone)	TEST EXAMPLE (Biocide mixture)
<i>Enterobacter aerogens</i> ATCC 13048	390	195
<i>Staphylococcus aureus</i> ATCC 6538	195	195
<i>Staphylococcus epidermis</i> ATCC 155	390	97
<i>Bacillus subtilis</i> ATCC 6984	390	195
<i>Saccharomyces cerevisiae</i> ATCC 9763	390	390
<i>Rhizopus oryzae</i> ATCC 10404	390	195
<i>Aspergillus niger</i> ATCC 9642	195	195

As can be shown from the above Table 2, a biocide mixture in which 3-isothiazolone and 25 wt% polyhexamethyleneguanidine phosphate are mixed using the synergistic index is much more effective in controlling microorganisms than is a biocide with a single component of 1.5 wt% 3-isothiazolone.

[TEST EXAMPLE 2 AND COMPARATIVE EXAMPLE 2]

In order to determine the killing time to kill microorganisms when the biocide mixture used in TEST EXAMPLE 1 and 1.5% 3-isothiazolone that was used in COMPARATIVE EXAMPLE 1, measurements were made of the immediate effectiveness, the durability, and the number of strains by taking a strain solution at times corresponding to 0 hours, 3 hours, 24 hours, 48 hours, 72 hours, and 96 hours after respectively putting in 50, 100, and 200 ppm of a biocide mixture and 1.5% 3-isothiazolone into the cooling water of a polymerization cooling tower having a microorganism concentration of about 104

CFU/ml., A solution in which a biocide was not added was used as a blank. The measured results on the strain reduction ratios according to the time when the biocide mixture in which 3-isothiazolone and polyhexamethyleneguanidine phosphate was mixed are presented using a synergistic index, as well as data
 5 for single component biocide of 1.5 wt% 3-isothiazolone, are represented in Table 3 and Table 4.

[Table 3]

Measured results on the reduction ratio of microorganisms according to the concentration of a biocide mixture verses time (units: CFU/ml):

10

		0	3 hours	24 hours	48 hours	72 hours	96 hours
Blank		13,000	> 13,000	> 13,000	> 13,000	> 13,000	> 13,000
Biocide mixture	50 ppm	13,000	210	140	90	40	50
	100 ppm	13,000	80	170	90	50	30
	200 ppm	13,000	50	90	50	40	50

[Table 4]

Measured results on the reduction ratio of microorganisms according to
 15 the concentration verses time of 1.5 wt% of 3-isothiazolone (units: CFU/ml):

		0	3 hours	24 hours	48 hours	72 hours	96 hours
Blank		13,000	> 13,000	> 13,000	> 13,000	> 13,000	> 13,000
1.5 wt% of isothiazolone	50 ppm	13,000	12,000	450	> 10,000	> 10,000	> 10,000
	100 ppm	13,000	11,400	370	6,250	> 10,000	> 10,000
	200 ppm	13,000	5,300	200	2,100	> 10,000	> 10,000

As can be shown in the above Table 3 and Table 4, the single compound 1.5 wt% of 3-isothiazolone biocide shows sterilizing capability after 24 hours, while the immediate effectiveness is low and there is also a secondary
 20 propagation of microorganisms after 48 hours. The biocide mixture of 3-isothiazolone and polyhexamethyleneguanidine phosphate achieves synergistic effects as seen in immediate effectiveness, durability, and superior sterilizing effects on microorganisms as compared to the single component biocide. Therefore, it can be shown that when 3-isothiazolone and
 25 polyhexamethyleneguanidine phosphate are mixed, the disadvantage of 3-isothiazolone with immediate effectiveness and the problems of

polyhexamethyleneguanidine phosphate associated with antibiotic spectrum are mutually compensated, and sterilizing capability is correlated with a synergistic effect index.

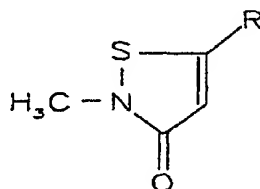
As described in the above, a biocide composition of the present invention can be effectively used in the wide control of microorganisms in various industrial fields such as water treatment, disinfectants, etc., since it has immediate effectiveness and durability, as well as applicability to a wide antibiotic spectrum. Furthermore, a biocide composition of the present invention can effectively control industrial water microorganism contamination and the actual living environment of the microorganisms since it is more effective in controlling the microorganisms and has greater immediate sterilizing capability than does a single compound biocide. A biocide composition of the present invention has an effect to lower the emergence frequency of a resistant strain as compared a single compound biocide, achieving this using a mixture of biocides having the different working mechanism.

While the present invention has been described in detail with reference to the preferred embodiments, those skilled in the art will appreciate that various modifications and substitutions can be made thereto without departing from the spirit and scope of the present invention as set forth in the appended claims.

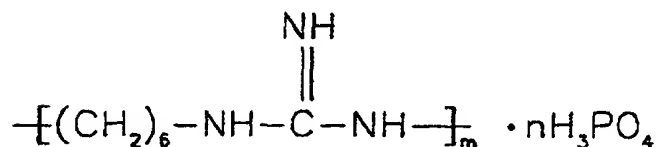
WHAT IS CLAIMED IS:

A biocide composition comprising 3-isothiazolone of the following General Formula 1 and polyhexamethyleneguanidine phosphate of the following General Formula 2:

5 [General Formula 1]



[General Formula 2]



10 where R is hydrogen or chlorine in the above General Formula 1, m is an integer from 4 to 7 and n is an integer from 1 to 14 in the above General Formula 2.

2. A biocide composition in accordance with claim 1 wherein, the mixing ratio of the above 3-isothiazolone and polyhexamethyleneguanidine phosphate
15 is a weight ratio of 1:1 to 1:65.

3. A biocide composition in accordance with claim 1 wherein, the above 3-isothiazolone is a mixture in which 3-isothiazolone having R of hydrogen and 3-isothiazolone having R of chlorine are mixed in a weight ratio of 1:20 to 20:1 and the mixing ratio of the above 3-isothiazolone and
20 polyhexamethyleneguanidine phosphate is from 1:1 to 1:4.

4. A biocide composition in accordance with claim 1 characterized in that before the use of the above biocide composition, it is mixed with a media selected from the group consisting of cooling water of an industrial process, disinfectant, paint, antiseptic for latex, additives for cosmetics, additives for
25 emulsion products, slime control chemicals for textile weaving, paper slime

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control agent, antiseptic for leather goods, and antiseptic for metal processing oil.

5. A sterilizing method of killing or restraining the growth of bacteria, fungi and/or algae by putting a biocide composition of claim 1 into the area that is contaminated by bacteria, fungi and/or algae.

5

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T05290 6267E860

PRIOR FOREIGN/PCT APPLICATION(S) FILED WITHIN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO SAID APPLICATION			
COUNTRY	APPLICATION NO.	DATE OF FILING (day, month, year)	DATE OF ISSUE (day, month, year)
Korea	Pat. 1998-49095	16/11/1998	
ALL FOREIGN APPLICATION(S), IF ANY, FILED MORE THAN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO SAID APPLICATION			

Claim for Benefit of Prior U.S. Provisional Application(s)

I hereby claim the benefit under Title 35, United States Code, § 119(e) of any United States provisional application(s) listed below:

Provisional Application Number	Filing Date

Claim for Benefit of Earlier U.S./PCT Application(s) under 35 U.S.C. 120
(complete this part only if this is a divisional, continuation or C-I-P application)

I hereby claim the benefit under Title 35, United States Code, § 120 of any United States application(s) or PCT international application(s) designating the United States of America that is/are listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior application(s) in the manner provided by the first paragraph of Title 35, United States Code § 112, 1 acknowledge the duty to disclose information as defined in Title 37, Code of Federal Regulations, § 1.56 which occurred between the filing date of the prior application(s) and the national or PCT international filing date of this application:

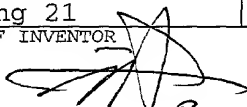

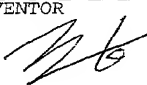
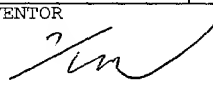
PCT/KR99/00687	16/11/1999	pending
(Application Serial No.)	(Filing Date)	(Status) (patented, pending, abandoned)

Power of Attorney

As a named inventor, I hereby appoint Dana M. Raymond, Reg. No. 18,540; Frederick C. Carver, Reg. No. 17,021; Francis J. Hone, Reg. No. 18,662; Joseph D. Garon, Reg. No. 20,420; Arthur S. Tenser, Reg. No. 18,839; Ronald B. Hildreth, Reg. No. 19,498; Thomas R. Nesbitt, Jr., Reg. No. 22,075; Robert Neuner, Reg. No. 24,316; Richard G. Berkley, Reg. No. 25,465; Richard S. Clark, Reg. No. 26,154; Bradley B. Geist, Reg. No. 27,551; James J. Maune, Reg. No. 26,946; John D. Mumane, Reg. No. 29,836; Henry Tang, Reg. No. 29,705; Robert C. Scheinfeld, Reg. No. 31,300; John A. Fogarty, Jr., Reg. No. 22,348; Louis S. Sorell, Reg. No. 32,439 and Rochelle K. Seide Reg. No. 32,300 of the firm of BAKER & BOTTS, L.L.P., with offices at 30 Rockefeller Plaza, New York, New York 10112, as attorneys to prosecute this application and to transact all business in the Patent and Trademark Office connected therewith

SEND CORRESPONDENCE TO: BAKER & BOTTS, L.L.P. 30 ROCKEFELLER PLAZA, NEW YORK, N.Y. 10112 CUSTOMER NUMBER 21003	DIRECT TELEPHONE CALLS TO: BAKER & BOTTS, L.L.P. 212) 705-5000
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I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon

1-02	FULL NAME OF SEVENTH INVENTOR	LAST NAME <u>CHOI</u>	FIRST NAME <u>Ki-Seung</u>	MIDDLE NAME	
	RESIDENCE & CITIZENSHIP	CITY <u>Uiwang-city,</u> <u>Kyungki-do</u>	STATE or FOREIGN COUNTRY Korea	COUNTRY OF CITIZENSHIP Korea <u>KRX</u>	
	POST OFFICE ADDRESS	POST OFFICE ADDRESS <u>Jindalrae Apt.103-302,</u> <u>Ojeon-dong 21</u>	CITY <u>Uiwang-city,</u> <u>Kyungki-do</u>	STATE or COUNTRY Korea	ZIP CODE 437-070
	DATE <u>5/19/01</u>	SIGNATURE OF INVENTOR 			
2-02	FULL NAME OF SEVENTH INVENTOR	LAST NAME <u>KIM</u>	FIRST NAME <u>Jin-Man</u>	MIDDLE NAME	
	RESIDENCE & CITIZENSHIP	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or FOREIGN COUNTRY Korea	COUNTRY OF CITIZENSHIP Korea <u>KRX</u>	
	POST OFFICE ADDRESS	POST OFFICE ADDRESS <u>Cheonrok Apt. 3-306,</u> <u>Yuljeon-dong,</u> <u>Changan-ku</u>	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or COUNTRY Korea	ZIP CODE 440-320
	DATE <u>5/19/01</u>	SIGNATURE OF INVENTOR 			
3-02	FULL NAME OF SEVENTH INVENTOR	LAST NAME <u>PARK</u>	FIRST NAME <u>Jeong-Ho</u>	MIDDLE NAME	
	RESIDENCE & CITIZENSHIP	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or FOREIGN COUNTRY Korea	COUNTRY OF CITIZENSHIP Korea <u>KRX</u>	
	POST OFFICE ADDRESS	POST OFFICE ADDRESS <u>Dongshin Apt. 201-1109,</u> <u>Jungja 1-dong,</u> <u>Changan-ku</u>	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or COUNTRY Korea	ZIP CODE 440-301
	DATE <u>5/19/01</u>	SIGNATURE OF INVENTOR 			
4-02	FULL NAME OF SEVENTH INVENTOR	LAST NAME <u>CHO</u>	FIRST NAME <u>Myung-Ho</u>	MIDDLE NAME	
	RESIDENCE & CITIZENSHIP	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or FOREIGN COUNTRY Korea	COUNTRY OF CITIZENSHIP Korea <u>KRX</u>	
	POST OFFICE ADDRESS	POST OFFICE ADDRESS <u>Samsung Apt. 1-805,</u> <u>Kwonseon-dong,</u> <u>Kwonseon-ku</u>	CITY <u>Suwon-city,</u> <u>Kyungki-do</u>	STATE or COUNTRY Korea	ZIP CODE 441-390
	DATE <u>5/19/01</u>	SIGNATURE OF INVENTOR 			

0033129-062901

[illegible]

[] Signature for ninth and subsequent joint inventors. Number of pages added _____

[] Signature by administrator(trix), executor(trix) or legal representative for deceased or incapacitated inventor.
Number of pages added _____

[] Signature for inventor who refuses to sign, or cannot be reached, by person authorized under 37 CFR 1.47.
Number of pages added _____

**COMBINED DECLARATION
AND POWER OF ATTORNEY**

(Original, Design, National Stage of PCT, Divisional, Continuation or C-1-P Application)

As a below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below next to my name; I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

BIOCIDE COMPOSITION AND STERILIZATION METHOD USING THE SAME

This declaration is of the following type:

- ☒ original
- ☐ design
- ☐ national stage of PCT.
- ☐ divisional
- ☐ continuation
- ☐ continuation-in-part (C-I-P)

the specification of which: (complete (a), (b), or (c))

- (a) ☒ is attached hereto.
- (b) ☐ was filed on as Application Serial No. _____ and was amended on (if applicable).
- (c) ☐ was described and claimed in PCT International Application No. _____ filed on _____ and was amended on (if applicable).

Acknowledgement of Review of Papers and Duty of Candor

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information which is material to the patentability of the subject matter claimed in this application in accordance with Title 37, Code of Federal Regulations § 1.56.

In compliance with this duty there is attached an information disclosure statement. 37 CFR 1.98.

Priority Claim

I hereby claim foreign priority benefits under Title 35, United States Code, § 119(a)-(d) of any foreign application(s) for patent or inventor's certificate or of any PCT International Application(s) designating at least one country other than the United States of America listed below and have also identified below any foreign applications) for patent or inventor's certificate or any PCT International Application(s) designating at least one country other than the United States of America filed by me on the same subject matter having a filing date before that of the application on which priority is claimed

(complete (d) or (e))

- (d) ☐ no such applications have been filed.
- (e) ☒ such applications have been filed as follows:

062990-062990